#### Genistein

Transformation of genistein by human intestinal bacteria

代謝実験

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# Anaerobic incubation with human intestinal bacterium, monitoring a metabolite by HPLC

A bacterium was inoculated into 2 ml of GAM broth. When turbidities (540 nm) of the bacterial suspension reached to 0.30±0.02 O.D., a 100-µl portion of the precultured bacteria was inoculated into 2 ml of GAM broth with genistein. After incubation, a 100-µl aliquot was removed and extracted three times with 200 µl of BuOH. After evaporation of the BuOH *in vacuo*, the residue was dissolved in 0.3 ml of MeOH. The MeOH solution was filtered through a 0.2-µm membrane filter, and a 10-µl portion was injected onto a column for HPLC analysis under the conditions described above. [Jin *et al.*, *Biol. Pharm. Bull.*, **31**, 1621-1625 (2008).]

### Preparation of 5-hydroxyeqoul by a human intestinal bacterial strain DZE

A bacterial suspension (50 ml) of strain DZE was inoculated to 0.8 1 GAM broth containing genistein (50 mg) and incubated at 37°C in an anaerobic incubator for 120 h. The reaction mixture was then extracted three times with ethyl acetate. The organic layer was evaporated under reduced pressure to give a residue. The residue was applied to a column of silica gel, which was eluted with a solvent system, CHCl<sub>3</sub>-MeOH (20:1),

to give 5-hydroxyequol (24 mg, 50% in yield). [Jin *et al.*, *Biol. Pharm. Bull.*, **31,** 1621-1625 (2008).]

## 5-Hydroxyequol

Amorphorus powder. EI-MS m/z: 258 [M]<sup>+</sup>. [ $\alpha$ ]<sup>23</sup><sub>D</sub> -9.0° (c=0.256, MeOH). This compound was identified by comparing the <sup>1</sup>H-NMR spectrum with that published. [Jin *et al.*, *Biol. Pharm. Bull.*, **31**, 1621-1625 (2008).]

## 参考文献

1) Jin J. S., Nishihata T., Kakiuchi N. and Hattori M.: Biotransformation of *C*-glucosylisoflavone puerarin to estrogenic (3*S*)-equol in co-culture of two human intestinal bacteria. *Biol. Pharm. Bull.*, **31**, 1621-1625 (2008).